UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, DC 20460

OFFICE OF
CHEMICAL SAFETY
AND POLLUTION

December 05, 2012

MEMORANDUM

SUBJECT: Efficacy Review for Hype-Wipe;
EPA Reg. No. 70590-1;
DP Barcode: D405415

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APPLICANT: Current Technologies, Inc.
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Formulation from the Label:

Active Ingredient(s):	% by wt.
Sodium Hypochlorite	0.94%
Inert Ingredients	99.06%
Total	100.00%

I. BACKGROUND:

The product, Hype-Wipe Disinfecting Towel with Bleach (EPA Reg. No. 70590-1), is an EPA-approved disinfectant towelette for use on hard non-porous surfaces in residential, commercial, and institutional environments. The applicant requested to amend the registration of the product to add disinfection claims for sporicidal activity against the organism *Clostridium difficile*. Studies were conducted at ATS Labs, located at 1285 Corporate Center Drive, Suite 110, in Eagan, MN 55121.

This data package contained a letter from the applicant to EPA dated September 07, 2012 (MRID 489337-00), three studies (MRID 489337-01 and 489337-03), Statement of No Data Confidentiality Claims for all studies, and the proposed label.

II. USE DIRECTIONS:

The product is designed for disinfecting hard non-porous environmental surfaces including baked enamel, plexiglass, glazed porcelain, linoleum, glass, plastic, glazed ceramic tile, laminated plastic counters, some floor types, and some grades of stainless steel.

Directions on the proposed label provide the following information regarding use of the product as a disinfectant:

(1) Remove all gross filth and heavy soil from surfaces to be disinfected. Gloves should be worn. (2) Open pouch, remove towel. Use towel and excess liquid to wipe surface. (3) Allow solution to remain wet on surface for at least two minutes to inactivate a broad range of micro-organisms. Leave wet four minutes to kill C. Diff spores. See front panel for specific pathogen kill times. (4) Rinsing: Some equipment / surfaces such as stainless steel may require rinsing: follow those specific manufacturers' directions.

Directions on the proposed label provide the following information regarding use of the product for special cleaning procedure before disinfecting against C. Diff spores:

Fecal matter/waste must be thoroughly cleaned from surfaces/objects before disinfection by application with a clean cloth, mop, and/or sponge saturated with the disinfectant product. Cleaning is to include vigorous wiping and/or scrubbing, until all visible soil is removed. Special attention is needed for high-touch surfaces. Surfaces in patient rooms are to be cleaned in an appropriate manner, such as from right to left to right, on horizontal surfaces, and top to bottom, on vertical surfaces, to minimize spreading or spores. Restrooms are to be cleaned last. Do not reuse soiled cloths.

III. AGENCY STANDARDS FOR PROPOSED CLAIMS:

Antimicrobial Products for Use on Hard Surfaces Using Pre-saturated or Impregnated Towelettes:

Towelette products represent a unique combination of antimicrobial chemical and applicator, pre-packaged as a unit in fixed proportions. As such, the complete product, as offered for sale, should be tested according to the directions for use to ensure the

product's effectiveness in treating hard surfaces. The standard test methods available for hard surface disinfectants and sanitizers, if followed exactly, would not closely simulate the way a towelette product is used. Agency guidelines recommend that a simulated-use test be conducted by modifying the standard test methods. Agency guidelines further recommend that instead of spraying the inoculated surface of the carrier, the product should be tested by wiping the surface of the carrier with the saturated towelette, and then subculturing the slides after a specified holding time. Sixty carriers must be tested with each of 3 product samples, representing 3 different product lots, one of which is at least 60 days old. The towelette should be removed from its container and subsequently handled with sterile gloves. One towelette should be used to wipe at least 10 inoculated slides. To support products labeled as "disinfectants," killing on 59 out of 60 carriers is required to provide effectiveness at the 95% confidence level. One carrier with a surface area equivalent to ten 1 x 1 inch carriers or ten carriers each with a surface area of 1 x 1 inch should be wiped using one towelette per carrier set (for a total of six towelettes and 60 carriers) per batch. The area of the towelette used for wiping should be rotated so as to expose a maximum amount of its surface in the course of wiping a set of slides. A detailed description of the wiping procedure, including the towelette folding and rotation process should be included in the test protocol and documented in the raw data and final report. The applicant should submit their towelette protocol to the Agency for review and approval prior to conducting the test.

Sporicidal Disinfectant against *Clostridium difficile*:

The Agency has established interim guidance for the efficacy evaluation of antimicrobial products (e.g., dilutable products, ready-to-use products, spray products, towelettes) that are labeled for use to treat hard, non-porous surfaces in healthcare settings contaminated with spores of *Clostridium difficile*. The effectiveness of such a product must be substantiated by data derived from one of the following four test methods: Most recent version (2006) of AOAC Method 966.04 (For the AOAC Method 966.04, testing should be conducted with two separate batches of product, using 30 carriers per batch for testing of registered sterilants; and three separate batches of product (one of which is at least 60 days old), using 60 carriers per batch for testing of hospital disinfectants. For the quantitative tests, the carrier number specified in the test method should be used): AOAC Sporicidal Activity of Disinfectants Test, Method I for *Clostridium sporogenes*; AOAC Method 2008.05: Quantitative Three Step Method (Efficacy of Liquid Sporicides Against Spores of *Bacillus subtilis* on a Hard Nonporous Surface); ASTM E 2414-05: Standard Test Method for Quantitative Sporicidal Three Step Method (TSM) to Determine Efficacy of Liquids, Liquid Sprays, and Vapor or Gases on Contaminated Carrier Surfaces; or ASTM E 2197-02: Standard Quantitative Carrier Test Method to Evaluate the Bactericidal, Fungicidal, Mycobactericidal, and Sporicidal Potencies of Liquid Chemical Germicides. Modifications to each test method will be necessary to specifically accommodate spores of *Clostridium difficile*. Because *Clostridium difficile* is an obligate anaerobe, testing should ensure adequate incubation conditions for the recovery of viable spores. The following toxigenic strains of *Clostridium difficile* may be used for testing: ATCC 700792, ATCC 43598, or ATCC 43599. All products must carry a pre-cleaning step, thus no organic soil should be added to the spore inoculum. Results must show a minimum 6 log reduction of viable spores for quantitative assessments or no positive carriers/tubes for quantitative assessments in 10 minutes or less. Control carrier counts must be greater than 10^6 spores/carrier.

IV. COMMENTS ON THE SUBMITTED EFFICACY STUDY:

1. MRID 489337-01 "Pre-Saturated Towelettes for Hard Surface Sporicidal Activity," Test Organism: *Clostridium difficile*- spore form (ATCC 43598) for product Hype-Wipe Disinfecting Towel with Bleach, by Anne Stemper. Study conducted at ATS Labs 1285 Corporate Center Drive Suite 110 Eagen, MN. 55121. Study completion date – July 31, 2012. Project Identification Number A13583.

The study was conducted against *Clostridium difficile*- spore form (ATCC 43598). One lot of the product Hype-Wipe disinfecting towel with bleach, Lot 041112, was tested using the provided ATS Laboratory Protocol No. CNT01061412.STOW.2 marked as proprietary information. The lot was ≥60 days aged. The product lot testing substance was received as a ready to use wipe. The active ingredient concentration stated in the study information section was 0.527%. The test organism was prepared by inoculation of five 10 mL tubes containing BHI broth with the test organism from a stock source. The tubes were incubated for 2 days at 35 – 37°C under anaerobic conditions followed by inoculating 500 µL per plate onto 80 CDC Anaerobic Blood agar plates. The plates were incubated for 7 days at 35 – 37°C under anaerobic conditions. Following incubation, 3.0 mL of sterile deionized water was added to each plate and each plate was gently scraped to harvest the growth. The suspension was removed, centrifuged, and resuspended in sterile deionized water twice prior to the final removal of the supernatant and resuspending the pellet in sterile deionized water. The culture was macerated to uniformity and stored at 2- 8°C for approximately two months prior to use. The spore purity was examined by Malachite green stain and microscopic analysis and found to be at a 92% spore to vegetative cell ratio. Glass slides (1 inch X 1 inch) were inoculated with 10 µL of culture that was uniformly spread over an approximate area of 1 inch X 1 inch and the inoculated slides were dried for 38 minutes at 35 -37°C with 50 - 51% relative humidity. One towelette was used to wipe 10 carriers by folding in half lengthwise twice and rolled up five times prior to use. The wiping procedure was done by exposing the maximum amount of the towelette surface area and passing it over the carrier surface back and forth twice for a total of four (4) passes for each inoculated carrier. The carriers were exposed for a 4 minutes contact period at 20°C with 55% relative humidity. Afterwards, each carrier was transferred to the primary subculture, 40 mL of Modified Fluid Thioglycollate Medium + 0.1% Sodium Thiosulfate + 0.1% Cholic Acid. Following the carriers were transfered into a secondary subculture, 40 mL of Modified Fluid Thioglycollate Medium + 0.1% Cholic Acid. A wetness test was performed for each carrier prior to subculturing and found to remain wet during the testing period. The plates were incubated for 48 ± 4 hours at 35 – 37°C under anaerobic conditions and stored at 2 – 8°C for two days prior to examination. All subculture vessels were incubated for 21 days at 35 – 37°C under anaerobic conditions prior to visual examination for the presence or absence of growth. Subcultures showing growth were subcultured, stained and/or biochemically assayed to confirm or rule out the presence of the test organism. Controls included purity, sterility, viability, HCl resistance, carrier population, and neutralization.

Note- For the HCl resistance control assay, four inoculated and dried carriers were transferred to vessels containing 40 mL of 2.5N HCl at room temperature and one carrier per time period was incubated for either 2 minutes, 5 minutes, 10 minutes, or 20 minutes prior to transferring each to primary vessels containing 40 mL of Modified Fluid Thioglycollate medium and secondary vessels containing 40 mL of Modified Fluid

Thioglycollate medium. The vessels were incubated with the test vessels. The acceptance criterion is to observe growth after at least 2 minutes of exposure.

2. MRID 489337-02 "Pre-Saturated Towelettes for Hard Surface Sporicidal Activity," Test Organism: *Clostridium difficile*- spore form (ATCC 43598) for product Hype-Wipe Disinfecting Towel with Bleach, by Anne Stemper. Study conducted at ATS Labs 1285 Corporate Center Drive Suite 110 Eagen, MN. 55121. Study completion date – August 28, 2012. Project Identification Number A13759.

The study was conducted against *Clostridium difficile*- spore form (ATCC 43598). Two lots of the product Hype-Wipe disinfecting towel with bleach, Lot 060112 and Lot 060512, were tested using the provided ATS Laboratory Protocol No. CNT01060412.STOW marked as proprietary Information. The product lot testing substance was received as a ready to use wipe. The active ingredient concentration stated in the study information section was 0.527% for Lot 060112 and 0.526% for Lot 060512. The test organism was prepared by inoculation of five 10 mL tubes containing BHI broth with the test organism from a stock source. The tubes were incubated for 2 days at 35 – 37°C under anaerobic conditions followed by inoculating 500 µL per plate onto 80 CDC Anaerobic Blood agar plates. The plates were incubated for 7 days at 35 – 37°C under anaerobic conditions. Following incubation, 3.0 mL of sterile deionized water was added to each plate and each plate was gently scraped to harvest the growth. The suspension was removed, centrifuged, and resuspended in sterile deionized water three times prior to the final removal of the supernatant and resuspending the pellet in sterile deionized water. The culture was macerated to uniformity and stored at 2- 8°C for approximately three months prior to use. The spore purity was examined by Malachite green stain and microscopic analysis and found to be at a 92% spore to vegetative cell ratio. Glass slides (1 inch X 1 inch) were inoculated with 10 µL of culture that was uniformly spread over an approximate area of 1 inch X 1 inch and the inoculated slides were dried for 38 minutes at 35 -37°C with 50% relative humidity. One towelette was used to wipe 10 carriers by folding in half lengthwise twice and rolled up five times prior to use. The wiping procedure was done by exposing the maximum amount of the towelette surface area and passing it over the carrier surface back and forth twice for a total of four (4) passes for each inoculated carrier. The carriers were exposed for a 4 minutes contact period at 20°C with 61% relative humidity. Afterwards, each carrier was transferred to the primary subculture, 40 mL of *C. diff* Broth + 0.1% Sodium Thiosulfate. Following, the carriers were transfered into a secondary subculture of 40 mL of C. *diff* Broth. A wetness test was performed for each carrier prior to subculturing and found to remain wet during the testing period. The plates were incubated for 48 ± 4 hours at 35 – 37°C under anaerobic conditions and stored at 2 – 8°C for two days prior to examination. All subculture vessels were incubated for 21 days at 35 – 37°C under anaerobic conditions prior to visual examination for the presence or absence of growth. Subcultures showing growth were subcultured, stained and/or biochemically assayed to confirm or rule out the presence of the test organism. Controls included purity, sterility, viability, HCl resistance, carrier population, and neutralization.

Note- For the HCl resistance control assay, four (4) inoculated and dried carriers were transferred to vessels containing 40 mL of 2.5N HCl at room temperature and one carrier per time period was incubated for either 2 minutes, 5 minutes, 10 minutes, or 20 minutes prior to transferring each to primary vessels containing 40 mL of Modified Fluid

Thioglycollate medium and secondary vessels containing 40 mL of Modified Fluid Thioglycollate medium. The vessels were incubated with the test vessels. The acceptance criterion is to observe growth after at least 2 minutes of exposure.

3. MRID 489337-03 "Wetness Determination for Towelette Products," Test Organism: *Clostridium difficile*- spore form (ATCC 43598) for product Hype-Wipe Disinfecting Towel with Bleach, by Becky Lien. Study conducted at ATS Labs 1285 Corporate Center Drive Suite 110 Eagen, MN. 55121. Study completion date – September 6, 2012. Project Identification Number A13934.

The study test substance was Hype-Wipe Disinfecting Towel with Bleach. Three lots (Lot 041112, Lot 060t t2, and Lot 0605t2) were examined using the provided ATS Labs Protocol No. CNT01071t t2.WET marked as proprietary information. One of the lots, Lot 041112, was ≥60 days aged. The product was received as a ready to use wipe. One towelette was used to wipe a glass (12 inch X 12 inch) carrier per lot of test substance. A video recorder was used to record the procedure from the start to finish. Prior to treatment, the carrier was weighed. The towelette was folded in half twice, once along the length and once along the width. The towelette was placed on the top left corner of the test carrier and wiped in an up and down motion, each stroke slightly overlapping the last, until the entire test carrier was completely covered for approximately 7 total strokes. A calibrated timer was initiated after the entire test surface was treated. The carrier was placed on the scale, the initial wet weight of carrier was taken, the carrier was allowed to be undisturbed for the exposure period of 4 minutes, and upon completion of the exposure period the final wet weight was taken. The test surface was wiped across a single sheet of unfolded cigarette paper immediately following final weighing to assist in visualization of wetness. Visual wetness of the cigarette paper was used to determine the presence or absence of carrier wetness. For the gravimetric wetness test, one towelette was used to wipe 10 glass slide carriers for each lot under ambient conditions. Prior to wiping, the towelette was folded in half lengthwise twice and rolled five times and each carrier in its empty aluminum weigh boat without a lid was weighed. Each carrier was wiped and immediately weighed. The exposure period of 4 minutes began once the carrier was wiped. Each carrier was weighed after the exposure period. Following, the carriers were dried for 30 minutes at approximately 102°C and cooled for at least one hour prior to being weighed. The acceptance criterion for this procedure is that the weight following the exposure time is greater than the dried weight for all carriers tested.

Note- For the Gravimetric Wetness Confirmation calculation of the percentage of moisture loss, the following calculation was applied:

Percent (%) Moisture Loss = $[1- (W_f - W_d) / (W_w - W_d)] \times 100$

Where:

W_d = Dried weight of treated slide

W_w = Weight of slide immediately following wiping

W_f = Final weight of slide following exposure time

V. RESULTS:

MRID Number	HCL Resistance Control 0 = No Growth + = Growth 1° = Primary Culture 2° = Secondary Culture								No. Exhibiting Growth/ Total No. Tested			Carrier Population (CFU/carrier) Log
	Minutes of exposure								Lot 041112 (≥60 days aged)	Lot 060112	Lot 060512	
	2		5		10		20		4-Minutes Exposure Period			
	1°	2°	1°	2°	1°	2°	1°	2°				
489337-01	0	+	+	+	+	+	0	+	1° = 0/60 2° = 0/60	---	---	3×10^6
489337-02	+	+	+	+	+	+	+	+	---	1° = 0/60 2° = 0/60	1° = 0/60 2° = 0/60	2.24×10^6

MRID 489337-03	Gravimetric Wetness (Avg. % Moisture Loss) (Pass/Fail)* 4 minutes exposure period	Visual Wetness Determination 4 minutes exposure period			
		Initial Weight of Carrier	Initial Wet Weight of Treated Carrier	Final Wet Weight of Treated Carrier	Visual Wetness on Cigarette Paper (Pass/Fail)†
Lot 041112	5.68% (Pass)	509.77 g.	510.63 g.	510.28 g.	Pass
Lot 060112	4.00% (Pass)	513.82 g.	514.97 g.	514.57 g.	Pass
Lot 060512	3.50% (Pass)	513.09 g.	514.05 g.	513.73 g.	Pass

*Passing results are demonstrated when the value for weight following the exposure time is greater than weight after drying.

†Passing results are demonstrated when visual wetness observed on the cigarette paper.

VI. CONCLUSIONS:

1. The submitted efficacy data does not support the use of the product, Hype-Wipe Disinfecting Towel with Bleach, as a Sporicide against the following microorganism on hard, non-porous surfaces for a 4-minute contact time:

Clostridium difficile MRIDs 489337-01 thru 489337-03

Results show no positive growth on all carriers/tubes tested in 10 minutes or less. Control carrier counts were greater than 10^6 spores/carrier. Growth was observed after at least 2 minutes exposure in the HCl resistance testing for the organism used in the examinations. Passing wetness determination for the contact period was demonstrated for all of the lots tested. Viability controls were positive for growth. Purity controls were reported as pure. Sterility controls did not show growth. Neutralization

controls did not demonstrate growth. **However, efficacy data were generated using an unapproved method for testing towelette products against *C. difficile*.**

VII. RECOMMENDATIONS:

1. The proposed label claims that the product, Hype-Wipe Disinfecting Towel with Bleach, is an effective disinfectant towelette against the following microorganism on hard, non-porous surfaces for a 4-minute contact time:

Clostridium difficile	ATCC 43598

The Agency does not feel comfortable accepting qualitative efficacy methods when testing towelette products against *Clostridium difficil*e, therefore these claims are **unacceptable**. In the absence of a validated quantitative method mimicking the actual use of towelette products, the registrants are encouraged to submit protocol for review before testing. For the moment, the Agency accepts quantitative efficacy data generated using ASTM E 2197 with expressed liquid (at LCL) from towelette. Contact time is based on wetness determination test.

2. The Agency recognizes the confusion on determining the appropriate test method for the evaluation of towelette products against C. *difficil*e spores. **In order to fully support claims against *C. difficile* spores, registrant must submit a confirmatory efficacy data, using ASTM E 2197 with expressed liquid (at LCL), on one lot of the towelette product Hype Wipe**.

3. These recommendations must be made on the proposed **LABEL**:

- Under "Special Cleaning Procedure Before Disinfecting Against C. Diff Spores" section, the pre cleaning step specifies cleaning with use of a "clean cloth, mop, and/or sponge saturated with the disinfectant product". This product is a towelette, therefore the statement must be changed to, "clean cloth, mop, and/or sponge saturated with a disinfectant product" or specify the disinfectant product that is in a ready to use liquid formulation that can be used by the application described.

- After *Clostridium difficil*e sporicide claims are approved, label must include these specific cleaning directions:

> **Personal Protection:** Wear appropriate barrier protection such as gloves, gowns, masks or eye covering.
> **Cleaning Procedure:** Fecal matter/waste must be thoroughly cleaned from surfaces/objects before disinfection by application with a clean cloth, mop, and/or sponge saturated with the disinfectant product. Cleaning is to include vigorous wiping and/or scrubbing, until all visible soil is removed. Special attention is needed for high-touch surfaces. Surfaces in patient rooms are to be cleaned in an appropriate manner, such as from right to left or left to right, on horizontal surfaces, and top to bottom, on vertical surfaces, to minimize spreading of the spores. Restrooms are to be cleaned last. Do not reuse soiled cloths.

Infectious Materials Disposal: Materials used in the cleaning process that may contain feces/wastes are to be disposed of immediately in accordance with local regulations for infectious materials disposal.

Data Submission: Information on the test system must be submitted including but not limited to the test design, spore production method, ATCC strain, spore titer, efficacy test method, neutralization study design and outcome, individual plate counts for treated and control carriers, and calculations including log reduction values. Any deviations to standard methods should be noted and supplied to the Agency. For the purpose of product registration, all studies are to be conducted following the Federal Insecticide, Fungicide and Rodenticide Act (FIFRA) Good Laboratory Practice Standards, 40 CFR Part 160, including media quality assessments and spore production.